Spring 2013

Continuous measurement of methane production before and after feeding in continuous cultures fed bermudagrass

K. M. Young

J. R. Burgess

C. T. McDonald

T. C. Jenkins

Continuous Measurement of Methane Production Before and After Feeding in Continuous Cultures Fed Bermudagrass

K.M Young*, J.R Burgess, C.T McDonald, T.C Jenkins;
Department of Animal and Veterinary Sciences. Clemson University, Clemson SC, United States

CLEMSON ANIMAL AND VETERINARY SCIENCES

ABSTRACT

The objective was to design a system that could detect gas concentrations continuously in ruminal continuous cultures to better assess changes in methane (CH_4) occurring before and after feeding. The custom-built system used CH_4 and CO_2 infrared sensors, and an O_2 infrared sensor. Thirty g of Tifton 85 bermudagrass harvested at 35 d of maturity was fed daily in two equal amounts at 0800 and 1600 h for three 7 d periods. For statistical analysis, hourly recordings were used between 0800 and 1600 h on days 5, 6 and 7. Data were analyzed by the Fit Model Procedure in JMP (SAS Institute) with time, day, and their interaction as main effects and period as random effect. Methane production increased (*P<0.01)* between 0800 and 1600 h (12.1, 12.6, 17.0, 21.9, 23.6, 26.4, 26.9, 27.1 and 28.1 mmol) when averaged across d 5-7. A decrease (*P*<0.01) in methane production on d *7* (26.1, 23.7, 15.4 mmol/d for d 5-7) is thought to result from repeated opening of the culture vessel to obtain samples. The novel gas analysis system developed for continuous cultures in this study successfully measured changes in CH_4 concentration before and after feeding that were comparable to previously reported values.

INTRODUCTION

Management Intensive Grazing Dairies (MiGDs) have increased in popularity in the southeast. This is mainly due to higher temperatures and therefore, a longer grazing season (Figure 1).

Increased time on pasture could play a role in methane production in the environment and agriculture accounts for 35-40% of the global anthropogenic methane production. (US EPA 2007)

Ruminal continuous cultures can be used to simulate digestion on pasture in a lab setting, and methane has been measured by analyzing spot samples via GC. But to date, no system has been developed to measure methane continuously in continuous cultures.

Figure 1. Dairy cattle in an intensive grazing system in GA

RESULTS

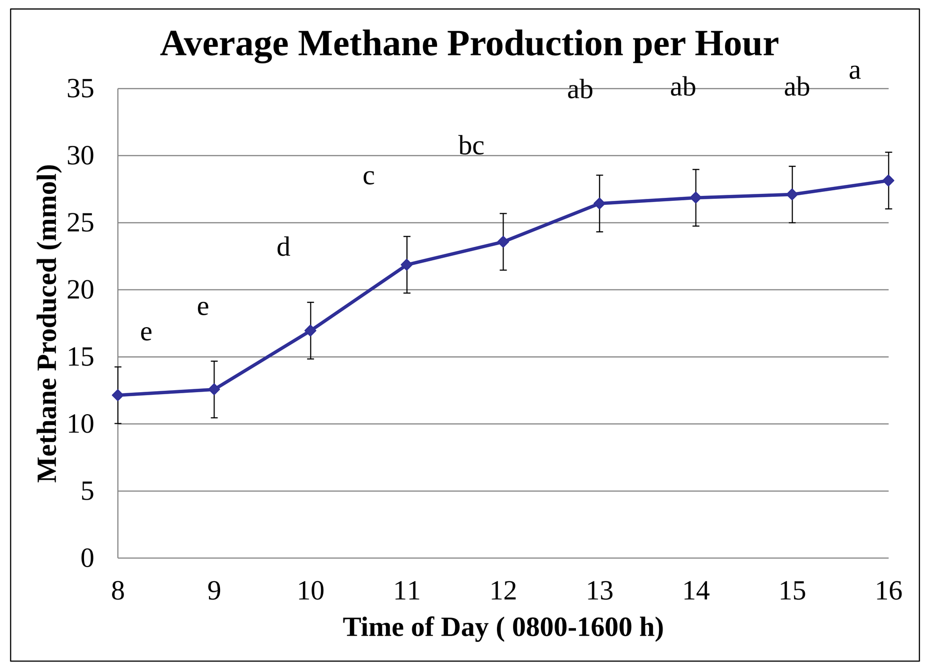

Figure 8. Methane production increased with time after feeding when cultures were fed at 0800 h. Means without a common superscript differ (*P<0.01).*

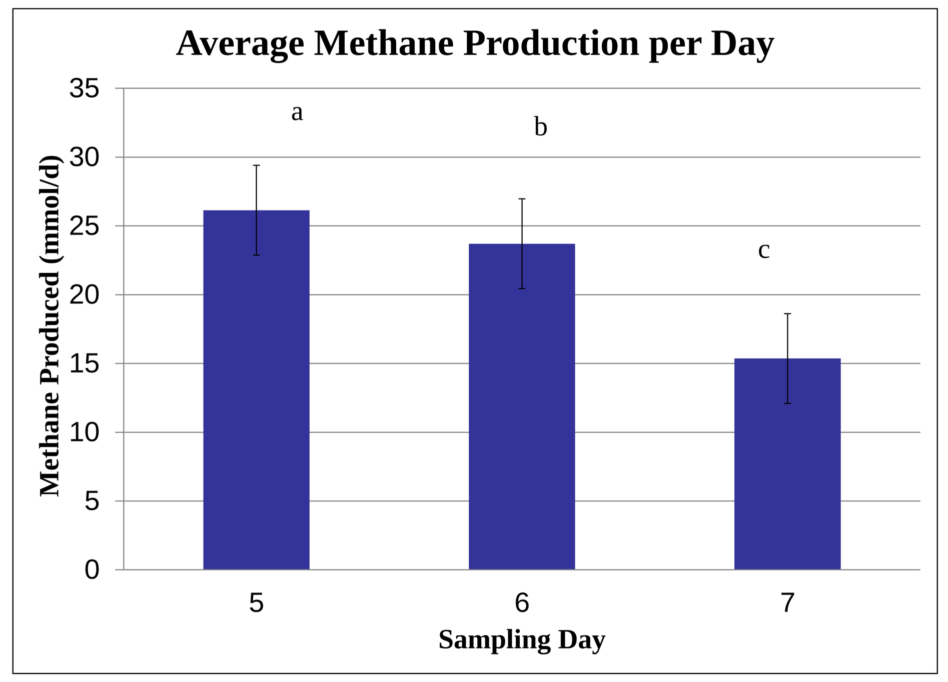

Figure 9. Methane production decreased with sampling day. Means without a common superscript differ (*P<0.01).*

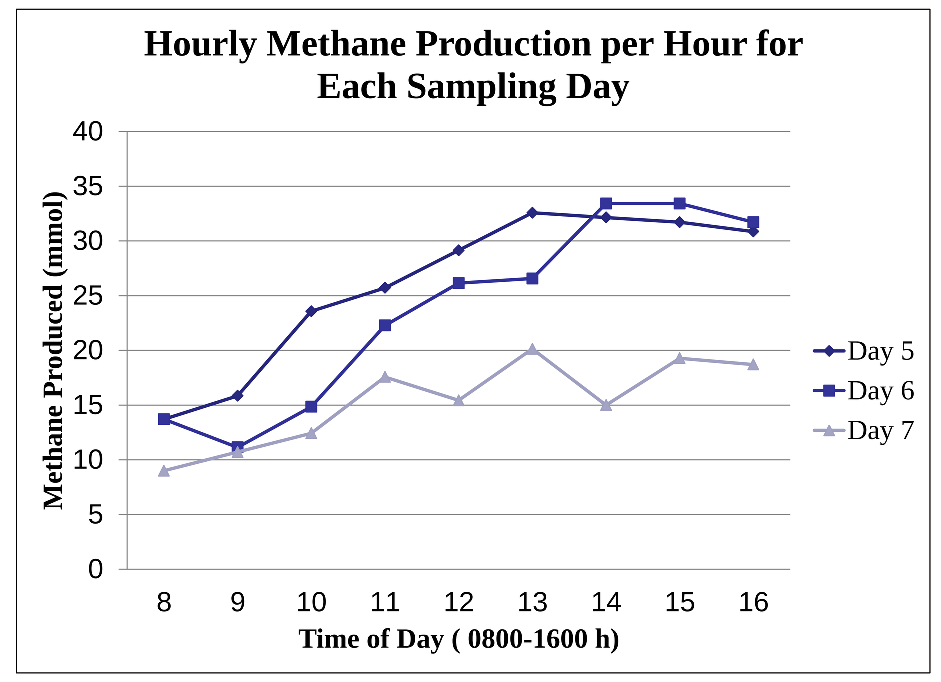

Figure 10. Hourly methane production separated by sampling day all followed a similar pattern of increasing with time after feeding.

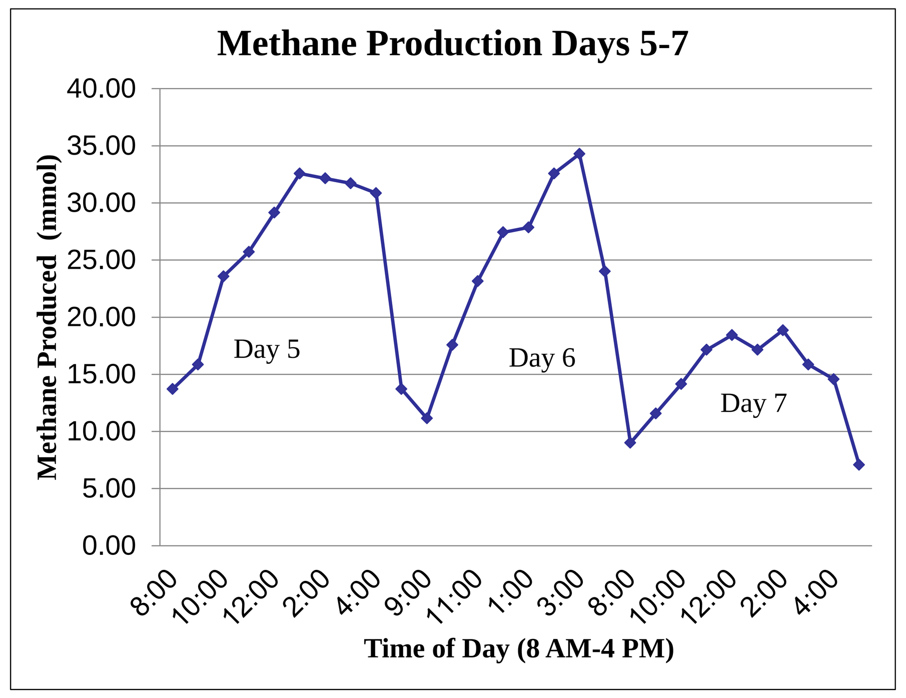

Figure 11. Hourly methane production over 3 days. Values were found to be lower in the morning before feeding and gradually increase after feeding. Cultures were fed at 0800 h and 1600 h.

Prediction of methane based on nutrient profile and DMI

Mills et al. (2003) equation: Methane(MJ/d) = 7.30 + 13.13 N (kg/d)+2.04 ADF (kg/d) + 0.33 Starch(kg/d)

***Modified Equation for continuous culture use:**

Methane (MJ/d) = 0.0057 + 13.13 N (kg/d) + 2.04 ADF (kg/d) +0.33 Starch(kg/d)

Methane(MJ/d) = 0.0057 + 13.13 *0.0007 + 2.04* 0.0071 +0.33*0.0012 = 0.03 MJ/d

Methane contains 0.891 MJ/mol

So 0.03 MJ/0.891MJ/mol *1000 = **33.8 mmol/d**

Nutrient input of 30 g bermudagrass:
Nitrogen: 0.0007 kg/d
ADF: 0.0071 kg/d
Starch: 0.0012 kg/d

*Since cultures are fed much less than a cow, we included 0.0057 MJ as the baseline production of methane instead of 7.30 MJ.

MATERIALS AND METHODS

Continuous culture care: 30 g of Tifton 85 bermudagrass was fed daily for three 7 d periods. pH and digester overflow were recorded twice daily right before feeding. Cultures (Figure 2) were kept under continuous flow of CO_2 at 20 mL/min and heated to 39 °C. Cultures were stirred continuously at a rate of 60 rpm in order to ensure proper stratification of feed as seen in the rumen. Buffer was pumped in continuously in order to maintain a 10-12% dilution rate in cultures.

Sample collection: Cultures were allowed four days for adjustment before taking gas readings. Readings were taken continuously every minute but only hourly readings between 0800 and 1600 h were used in the statistical analysis.

Setup Design: The CH_4 (Figure 3) and CO_2 (Figure 4) infared sensors (Edinburgh Instruments, OEM Gas Sensors , Great Britain) and an O_2 infared sensor (Figure 5) were calibrated with a zero and span gas prior to use. The three sensors are mounted on a stainless steel box (Figure 6) with an air pump to pull headspace sample through and a signal processor connected to a computer to record readings. Sample is pulled through silica tubing (Figure 7) of 3.18 mm ID and 6.35 mm OD (DOW Corning corporation, Midland, MI) through one of eight two-way pinch valves (Valcour Scientific, Springfield NJ) to allow sampling of one culture at a time. After sample is pulled through and a reading is taken, it is returned to the culture. A data acquisitioner is mounted on the box to measure voltage readings (0-5 V) to convert to gas percentages, which are captured on a spreadsheet.

Statistical Analysis : Data were analyzed using the Fit Model Procedure in JMP (SAS Institute) with time and day and their interaction as main effects and period as random effect. Time*day interactions were generated using Fisher's LSD Test.

Figure 2. Continuous culture complete with overflow port , stir rod, water pocket for heating, CO_2 and buffer pump.

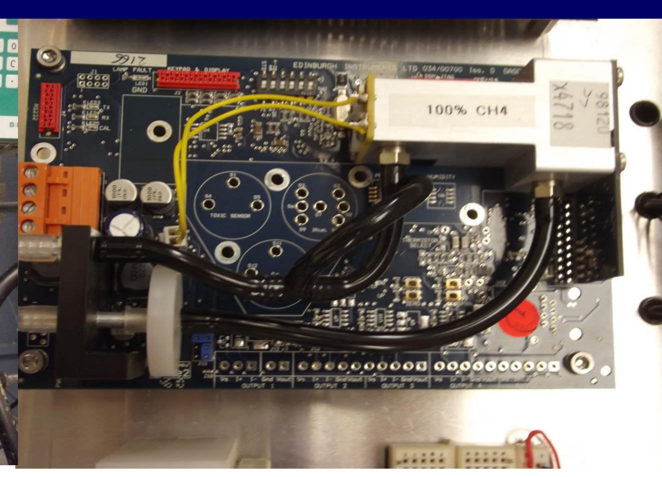

Figure 3. Methane sensor (Edinburgh Instruments, OEM Gas Sensors, Great Britain)

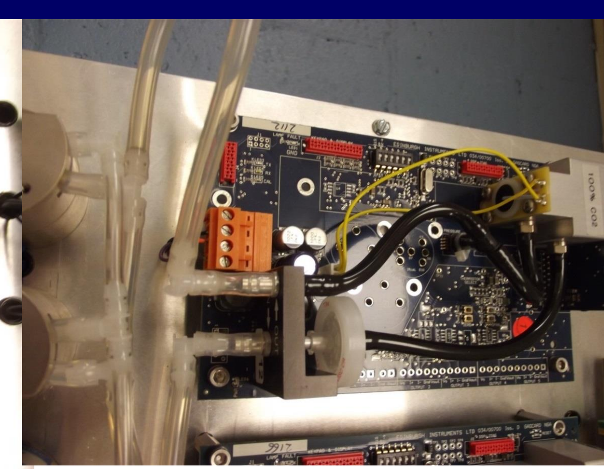

Figure 4. Carbon dioxide sensor (Edinburgh Instruments, OEM Gas Sensors, Great Britain)

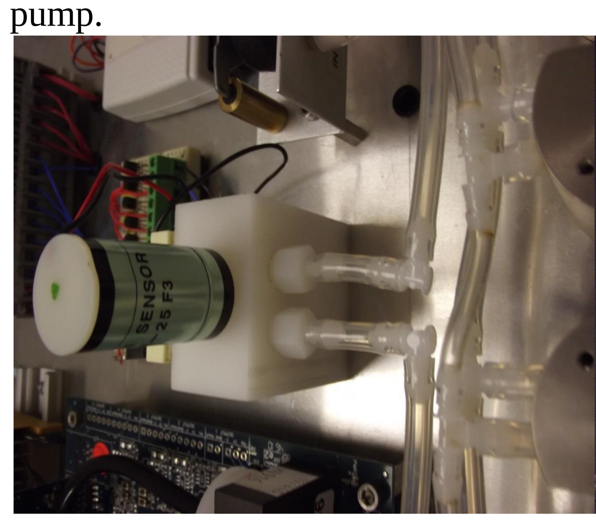

Figure 5. Oxygen sensor (KE Series, Figaro, USA Inc. Arlington Heights, IL)

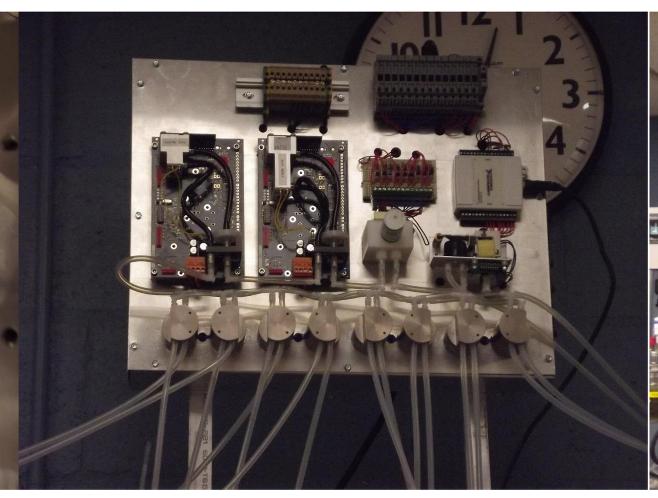

Figure 6. Stainless steel box with 8 two –way pinch valves and air pump, CH_4 , CO_2 and O_2 sensors, and data acquisitioner.

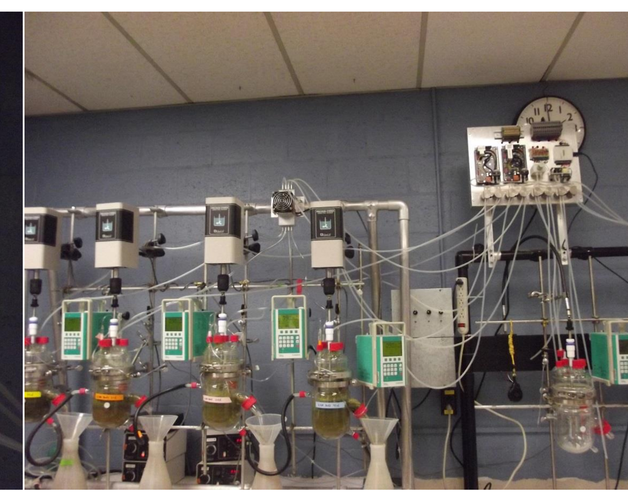

Figure 7. Continuous culture setup connected with gas sensor system by silica tubing.

DISCUSSION / CONCLUSIONS

In summary, The novel gas system **was effective in picking up differences in methane production before and after feedings, and methane production increases after feeding are likely due to increased substrates for methanogenesis**. However, the lower average daily methane production seen on day 7 implies that **increased opening of the culture vessel for sample collection can result in lower methane readings on those days likely due to air contamination**. Methane production predicted (33.8 mmol) from the equation by Mills et al. (2003) adapted for continuous culture use **was comparable to the methane values measured several hours after feeding (26.9, 27.1. 28.1 mmol)**. Since this equation uses the nutrient profile in feed to predict methane, it is assumed that values recorded several hours after feeding are most comparable since this is typically when methane production peaks after feeding. On the whole, this new setup appears to be promising but one needs to keep in mind the effects of increased sampling.

REFERENCES

- Mills, J. A. N., E. Kebreab, C.M Yates, L.A Crompton, S.B Cammell, M.S Dhanoa, and J. France. 2003. Alternative approaches to predicting methane emissions from dairy cows. Journal of Animal Science. 81(12):3141-3150
- US EPA. 2007. Inventory of U.S Greenhouse gas Emissions and Sinks: 1990-2005. EPA Report 430 R-06-002. http://www.epa.gov/climatechange/emissions/downloads06/07CR.pdf